

NITRITES AND NITROSAMINES IN OUR ENVIRONMENT: AN UPDATE¹

AARON E. WASSERMAN and IVAN A. WOLFF

*Agricultural Research, Science and Education Administration
U.S. Department of Agriculture
Eastern Regional Research Center
Philadelphia, Pennsylvania 19118*

INTRODUCTION

The difficulty of providing in a short time period an adequate status summary covering our environmental exposure to nitrites and nitrosamines and their health significance is evident from the information explosion shown in Fig. 3.1. We see a continuing acceleration of research effort in the field which now results in publication of a few hundred papers each year on various aspects of the subject. So we'll be able to provide an overview only of selected topics of greatest current interest.

The debate on the potential health hazards of nitrites and nitrosamines has by no means been restricted to the technical forums of scientific publications and gatherings. The popular media have become involved and emotions have often run high in discussions of the issues (Fig. 3.2). The plan for prompt ban of nitrite by some consumer groups has been strongly countered by industry and its associations who have cited a long record of apparent safety of established meat processing practices, as well as a response in the form of altered processing procedures designed to minimize nitrosamine formation. The stance of the regulatory agencies is governed to some extent by the proclivities of their current leaders who guide and direct regulatory policy within established law, and this has varied with the administration in office. Meanwhile, public research groups have laboriously sought the facts through laboratory research, a fruitful but slow and time-consuming process. Currently, superimposed on this picture are two additional groups which

¹This chapter includes data up to the period of early 1979.

EXPANSION OF RESEARCH ON NITROSATION AND NITROSAMINES

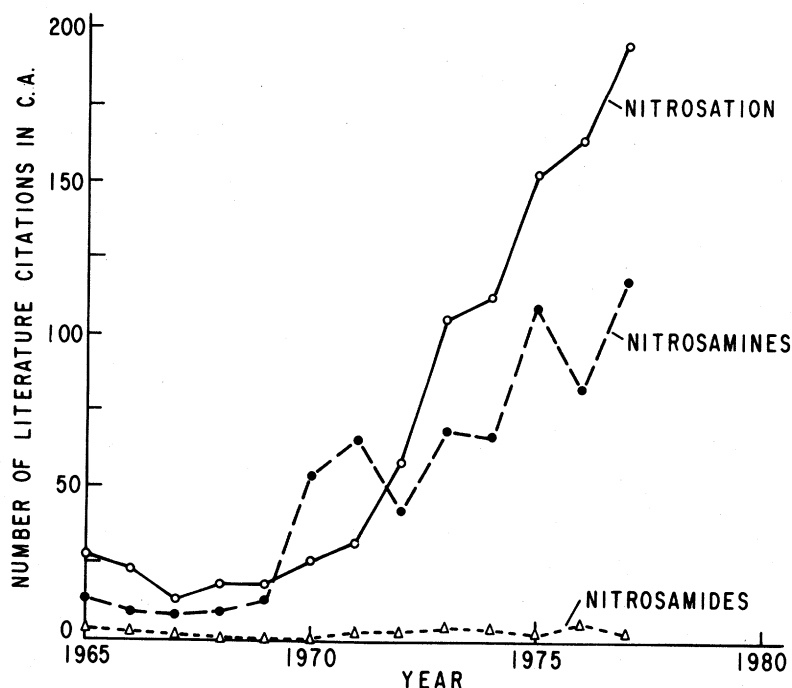


FIG. 3.1. INFORMATION EXPLOSION ON NITROSATION AND NITROSAMINES
Cited in Chemical Abstracts.

have become greatly involved—the Congress, which has considered legislating *against* a nitrite ban, and jurists who have been called upon to settle certain questions brought before the Courts, such as whether nitrite has prior sanction as a food preservation additive. So the situation on nitrites and nitrosamines is scientifically and socioeconomically complex.

From 1956 when the carcinogenicity of nitrosamines to experimental animals was initially reported until about 1970 and just after, research effort was focused on preformed nitrosamines in food, mainly in cured meat products. Since that time greatly improved analytical methodology has provided greater sensitivity and better procedures for determining nitrosamines. This has led to finding nitrosamines fairly ubiquitously in the environment: in foods, nonfood materials, the air, water, and in the human body. In addition, nitrite itself has been reported to be

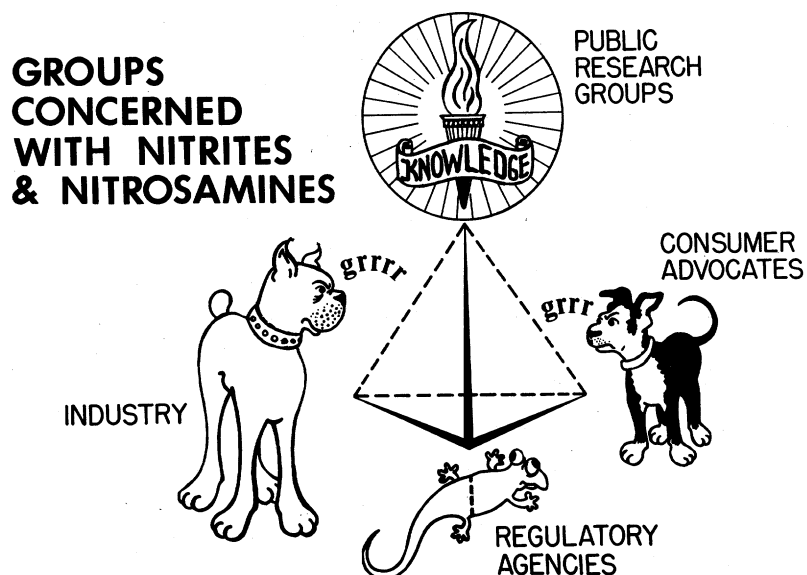


FIG. 3.2. GROUPS CONCERNED WITH NITRITES AND NITROSAMINES

carcinogenic to rats. As a consequence, much consideration is being given to risks and hazards to humans and to potential economic dislocations that might result from changes in United States meat curing procedures. These are the various areas that we'll try to summarize briefly.

IMPROVEMENTS IN METHODOLOGY

Nitrosamines and their precursors have been identified recently under conditions of increased instrumental sensitivity such that another dimension has been added to the carcinogenesis problem. Advances in analytical technology have permitted the identification and quantification of lower concentrations of nitrosamines. A new detector, the Thermal Energy Analyzer (TEA) is sensitive to 0.1 ng of a nitrosamine. Selectivity is enhanced because the detector monitors the chemiluminescence emitted when NO_2 , produced by oxidation of NO released from pyrolyzed nitrosamine, decays from the excited to the ground state. The increased sensitivity, however, has introduced another problem by reducing the quantifiable concentration of nitrosamine below the range of the mass spectrometer, which is needed for confirmation of identity. Tentative confirmation of the presence of nitrosamines can be obtained by the method of Doerr and Fiddler (1977) (Fig. 3.3) in which several μl of a sample, shown to contain nitrosamines, are exposed to UV

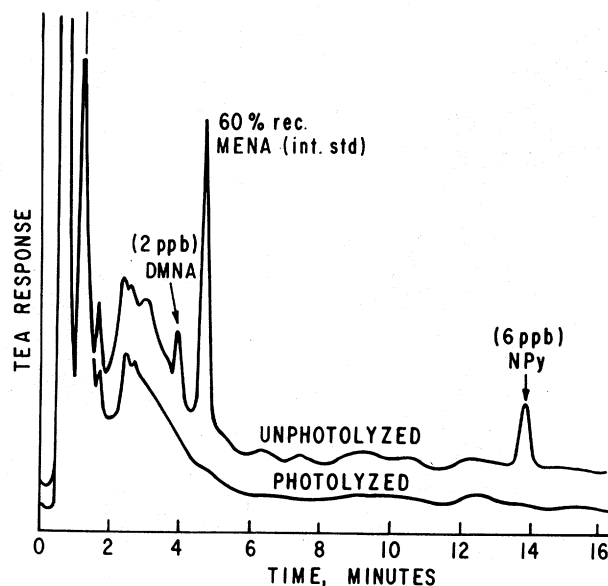


FIG. 3.3. EFFECT OF UV PHOTOLYSIS ON THE NITROSAMINES PRESENT IN AN EXTRACT OF FRIED BACON

at 366 nm for 2 h in a melting point capillary tube. When the sample is passed through the gas chromatograph-TEA detector again, nitrosamine peaks have disappeared. This serves as a good presumptive test and reduces the number of samples that need to be confirmed by mass spectrometry.

NITROSAMINES IN FOODS

Ongoing surveys of food products in several countries continue to report nitrosamines in meats, cheese, fish and prepared dinners. The levels of nitrosamines found, however, are lower than those reported in previous years. The use of the tandem gas chromatograph-TEA has permitted the detection of these compounds at concentrations less than 1 $\mu\text{g}/\text{kg}$ in products similar to those in which nitrosamine had not been found previously or had been found only in concentrations of more than 1 ppb (Gough 1978). In a series of 50 fried bacon samples, Gough *et al.* (1978) found 1–20 $\mu\text{g}/\text{kg}$ N-nitrosopyrrolidine (NPYR); N-nitrosopiperidine (NPIP), which had not been reported consistently in bacon, was now found in all samples at a concentration of 0.08 to 0.25 $\mu\text{g}/\text{kg}$ (or 80–250

ppt). Other meat products analyzed by Gough *et al.* (1978) contained several volatile nitrosamines, each in 0.1–1.0 $\mu\text{g/kg}$ concentration. These authors also analyzed a number of prepared meals and estimated the total weekly intake of volatile nitrosamines in English food (Table 3.1). The likely intake of dialkyl and heterocyclic nitrosamines was of the order of 1 μg and 3 $\mu\text{g/week}$, respectively. Stephany *et al.* (1976) found less than 5 $\mu\text{g/kg}$ of N-nitrosodimethylamine (NDMA), N-nitrosodiethylamine (NDEA), or N-nitrosodibutylamine (NDBA) in any one of the 36 samples of Dutch cured meat products examined; 50% of the samples contained less than 0.4 $\mu\text{g/kg}$ NDMA, NDEA, NDBA, NPYR, or NPIP.

TABLE 3.1
ESTIMATED WEEKLY INTAKE OF VOLATILE NITROSAMINES FROM
UK FOODS

Food	Consumption (kg per week) per person)	Dialkyl Nitrosamines		Heterocyclic Nitrosamines	
		Average concentration (μg per kg)	Intake (μg per week)	Average concentration (μg per kg)	Intake (μg per week)
Cured meats	0.34	1.0	0.34	8	2.7
Fish	0.14	0.2	0.03	—	—
Cheese	0.10	0.4	0.04	—	—
All other foods	9.62	0.06	0.58	—	—
Totals	10.20		0.99		2.7

Gough, *et al.* (1978).

To measure such low levels of nitrosamines, both groups of investigators used larger samples of meat than are commonly analyzed in the United States. The larger samples, however, contribute more interfering compounds that have to be removed. If the presence of low concentrations of nitrosamines becomes a critical regulatory factor, particularly at the international level, differences in samples size will have to be reconciled.

Although no pertinent scientific publication has appeared at the time this chapter was prepared, press releases reported the presence of NDMA in beer. Originally detected by Spiegelhalder *et al.* (1979) in Germany, scientists in Great Britain, Canada, and United States have confirmed the presence of NDMA, at concentrations up to 5 $\mu\text{g/L}$, in this fermented product. The origin of the nitrosamine has not been indicated but the malted barley may be the source. Sprouting could liberate the secondary amine, which could in turn react with nitrogen oxides in the gases present during the drying process.

NITROSAMINES IN NONFOOD SOURCES

NDMA was reported in six herbicide formulations in concentrations ranging from 0.3 to 640 mg/kg (Ross *et al.* 1977). These products were prepared as dimethylamine salts, which probably reacted with the sodium nitrite used as a rust inhibitor in the containers. A seventh herbicide, made with nitric acid followed by a reaction with dipropylamine, contained 154 mg/kg N-nitrosodipropylamine. The various companies involved have since taken steps to eliminate the formation of nitrosamines in their products.

Metal cutting oils were also a source of nitrosamines. Almost all of these products are synthetic, containing up to 45% triethanolamine and 18% NaNO₂. Eight brands, analyzed by Fan *et al.* (1977b), contained 0.02 to 2.99% N-nitrosodiethanolamine (NDELA). Triethanolamine is also used as an emulsifier in toiletry preparations such as cosmetics, lotions, and shampoos. Fan *et al.* (1977a) reported <10 to 40 mg/kg NDELA in 7 cosmetic preparations and <10 to 280 µg/L in 10 shampoos. Use of cosmetics may result in exposure to 50–100 µg/kg NDELA daily. The source of NDELA is not known, but it is possible that triethanolamine degrades to diethanolamine, which reacts with nitrogen oxides in the air.

The presence of NDMA has been reported in a tannery (Fajen *et al.* 1978). Dimethylamine sulfate is used in some tanneries as a component of a formulation in a bath for unhairing hides; it was found to be contaminated with NDMA, but at concentrations too low to account for the amount of NDMA found in the air around the unhairing operation (up to 36 µg/m³). Bailey *et al.* (1978) speculated that a reservoir of NDMA had accumulated on the surfaces of walls and equipment in the building and was bleeding into the atmosphere.

Nitrosamines have been reported in the air, particularly at point sources where either the nitroso compound or the precursor amine occurs in high concentrations. At Baltimore, Md., Belle, W. Va. (Fine *et al.* 1976), and near Boston, Mass. (Bailey *et al.* 1978), NDMA has been found in the atmosphere. Bretschneider and Matz (1976) also found NDMA in air associated with manufacturing activities in Germany.

Formation of nitrosamines in the atmosphere is possible if free amines react with nitrogen oxides, especially where a high SO₂ concentration produces acidic conditions. Photodecomposition of nitrosamines in the atmosphere by UV irradiation, however, appears to counterbalance the formation.

In tests of water from the Mississippi River a number of peaks detected by TEA were reported as "unidentified" nitrosamines (Fine *et al.* 1976); one peak, originally identified as nitrosoatrazine, was subsequently found to be ethylene glycol dinitrate. Fiddler *et al.* (1977) found

up to 0.2 $\mu\text{g/L}$ NDMA and NDEA in water that had been deionized by passage through certain ion exchange resins.

IN VIVO FORMATION

Evidence for the presence of nitrosamines in human body fluids and the potential for their formation *in vivo* is beginning to accumulate. Saliva has been found to contain 8–12 mg/L nitrite as a normal constituent. Ingestion of vegetables high in nitrate, however, results in a large increase in the nitrite concentration in the saliva—to hundreds of mg/L in a few hours. Purslane, a leafy salad green, for example (Fig. 3.4), induced high salivary nitrite for a period of several hours, as did cucumber and lettuce (Stephany and Schuller 1978) and celery juice (Tannenbaum *et al.* 1976). Since saliva obtained by cannulating the salivary glands contained no nitrite—only nitrate—the source of salivary nitrite is the reduction of nitrate to nitrite by bacteria in the mouth (Tannenbaum *et al.* 1976), and the resulting nitrite may react with amines in the gastric tract to form nitrosamines. High concentration of nitrate in the drinking water has been postulated to induce stomach cancer in some areas of Colombia, South America (Cuello *et al.* 1976), and a statistical correlation has been reported between high nitrate fertilization of crops and gastric cancer in Chile (Zaldivar 1977). In Iran, however, large amounts of nitrate in the water could not be related to the high incidence of esophageal cancer in which nitrosamines were thought to be the causal factor (Bogovski 1976).

In vivo formation of nitrite and nitrate occurs in the intestinal tract, according to Tannenbaum *et al.* (1978). Nitrification of ammonia, produced presumably by deamination of amino acids from organic nitrogen compounds, occurs in the upper part of the small intestine, with the formation of nitrite. Nitrate accumulates in the anaerobic atmosphere of the large intestine. Some of the nitrite and nitrate may be absorbed through the intestinal walls; nitrite is destroyed in the blood or reacts with hemoglobin, while some of the nitrate is recycled to the saliva and the remainder is excreted.

Dimethylamine, pyrrolidine, and other amines occur normally in the urine, as well as in the colon. Nitrate is also a normal urinary constituent, and in the presence of nitrate-reducing bacteria nitrosamine formation is possible. The urine of patients with urinary tract infections contained NDMA (Radomski and Hearn 1976), as did the urine of patients with cystitis, bladder cancer, and bilharzial infections of the bladder (Hicks *et al.* 1978).

Varghese *et al.* (1978) and Wang *et al.* (1978) have found a number of nitrosamines in normal human feces: 0.22–1.49 $\mu\text{g/kg}$ NDMA, 0.18–13.3 $\mu\text{g/kg}$ NDEA, 0.31–0.78 $\mu\text{g/kg}$ NPYR (in 3 of 12 people),

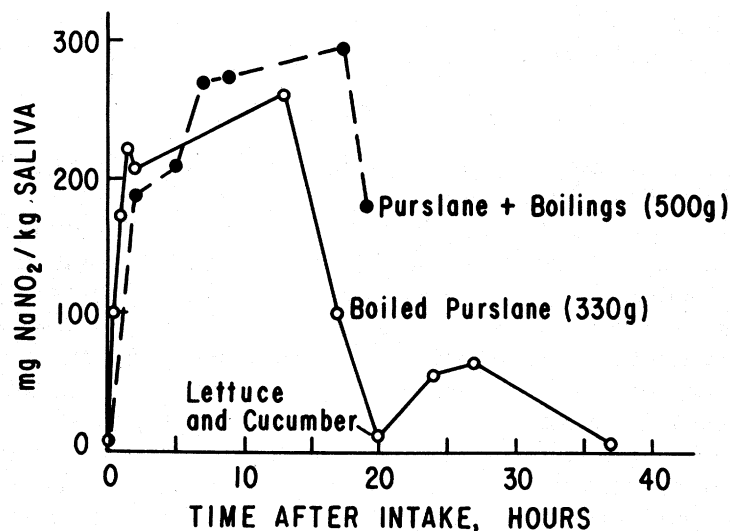


FIG. 3.4. EFFECT OF INGESTION OF VEGETABLES CONTAINING HIGH CONCENTRATIONS OF NITRATE ON THE NITRITE CONCENTRATION IN THE SALIVA

0.07–1.25 $\mu\text{g/kg}$ nitrosomorpholine (NMOR), and 0.12 and 0.87 $\mu\text{g/kg}$ N-nitrosodibutylamine (in 2 of 12 people). No explanation for the source of these compounds was given, but the authors postulate that they may be formed in the lower gastrointestinal tract.

Nitrosation occurs in the human stomach. Walters (1976) fed 14 volunteers milk, egg, and luncheon meat, then analyzed the stomach contents at various times. NPIP (0.3–0.5 $\mu\text{g/kg}$) was found in four subjects within 60 min; three were smokers, one was not. In another study, Fine *et al.* (1977) fed a volunteer bacon, spinach, tomato, bread, and beer. They examined the blood, instead of the stomach contents, and found no NPYR although it was present in the meal. However, they did detect NDMA in the blood, at a lower concentration than it appeared in the food.

In our laboratory we surveyed the fasting gastric contents of 35 patients, some before and after pentagastrin or histamine stimulation, in a preliminary study of the potential for nitrosation in the human stomach (Lakritz *et al.* 1978). Of 57 samples, 7 contained confirmable levels of nitrosamines (Table 3.2). Several samples contained “apparent” nitrosamines at levels too low to confirm.

We demonstrated the presence of NDMA in human blood and, in the limited number of samples tested, this nitrosamine appears to be a

TABLE 3.2.
NITROSAMINES IN HUMAN GASTRIC CONTENTS

Patient	Nitrosamine		NO ₂	NO ₃	pH	Diagnosis
	Compound	μg/g	mg/kg			
1	NDEA	30	69		1.6	Duodenal ulcer
	NDEA ¹	26	76		1.5	
2	NDEA	5			2.2	Marginal ulcer
3	NDEA	23			2.9	Marginal ulcer & cirrhosis of liver
4	NPYR ¹	6			6.4-7.5	Atrophic gastritis
5	NDMA	2	0	26	2.9	Not known
6	NDMA ¹	2			1.9	Gastritis, Duodenitis

¹These samples were obtained following gastric secretion stimulation with histamine or pentagastrin. Lakritz (1978).

normal constituent. In the blood of 37 of 38 people—male and female adults—we found NDMA in concentrations of 0.4 to 0.8 μg/L (Lakritz *et al.* unpublished). The source of the NDMA is not known at this time.

RISK

In all discussions of the risk of tumor formation in humans there has been an implication that the carcinogenic potential is the same for all nitrosamines. Reports in the literature, however, indicate that some are strong carcinogens (NDMA, NDEA), while others are weaker carcinogens (NPIP, nitrososarcosine (NSAR)), as shown by their behavior in animals. Archer and Wishnok (1977) presented interesting calculations on NDMA, NDEA, and NPYR, based on data from Druckery and his coworkers (1967) and Preussmann *et al.* (1976). The mean minimum doses of NDMA and NDEA that induced tumors in rats were 6 to 7 times less than that of NPYR, on a molar basis. The relative risk of these compounds can be obtained from the relative mole potency and from the concentration of nitrosamine in a food. Thus, at typical nitrosamine levels in processed meat (Table 3.3) NDMA is 5 or 6 times more dangerous than NPYR, and a concentration of, for example, 5 ppb NDMA in bacon should be of greater concern than the same amount of NPYR, if values obtained in test animals can be extrapolated to man.

While numerous animal studies have shown that tumor formation occurs following ingestion of nitrosamines, the studies have been criticized for the high concentrations used, since they may affect normal metabolic pathways or overwhelm the DNA repair mechanism that is currently under investigation (Magee 1976). Although Food and Drug Administration Commissioner Kennedy (1978) has discounted the

TABLE 3.3
NITROSAMINES IN PROCESSED MEAT

Nitrosamine	Typical Values				Relative Risk/g Food	
	High		Low		High Intake	Low Intake
	ppb	mol ¹	ppb	mol		
NDMA	25	0.34	3	0.04	48	6
NDEA	12	0.12	2	0.02	14	2
NPYR	50	0.50	5	0.05	10	1

¹mol $\times 10^9$ /g food.
Archer and Wishnok (1977).

threshold, or no response, dose for carcinogens, several recent studies showed that nitrosamines at some concentration in the diet induced no significant increase in tumors in rats. Olsen and Meyer (1976) fed 2 generations of rats a semi-synthetic diet containing 45% canned, cured meat as sole protein source (Table 3.4). Even with the meat cured with 4000 ppm nitrite and containing NDMA, no significant increase in tumors was found. A 3-year Canadian study (unpublished), just completed, involved feeding rats fried commercial bacon at 25% of their diet. The bacon was cured with 150 ppm nitrite and contained 39 ppb nitrosamines. No significant increase in tumors occurred. Preussmann *et al.* (1976) incorporated 10, 3, 1.0, or 0.3 mg NPYR/kg body weight in the daily drinking water of groups of rats (Table 3.5). Tumor formation was found at the three highest levels, but at 0.3 mg/kg the number of tumors was not significantly greater than in the controls. The "no effect" dose corresponds to 5 ppm in the diet. The latency period for tumor induction also increased with decreasing doses of NPYR, so it is possible that at low levels of nitrosamine the latency period could extend beyond the life span of the animal and no tumor would be formed.

Mantel (1978), at the request of the American Meat Institute, analyzed Preussmann's data and calculated a "safe" level of NPYR in bacon to be 24.8 ppb for the daily consumer, with a risk of less than one in a million over the lifetime.

NITRITE AS A CARCINOGEN

Although nitrite had been fed to animals in high concentrations as controls in many studies in the past, it produced no indication of a carcinogenic effect. Newberne (1978), however, in a study involving more than 2000 rats fed several diets containing various levels of nitrite,

TABLE 3.4
TUMOR FORMATION IN RAT-FEEDING¹ STUDY

Group	Nitrite Added mg/kg	Rats with Tumors		
		Total	Benign	Malignant
1	0	76	62	24
2	0	147	124	44
3	0	79	68	22
4	200	78	70	22
5	1000	78	67	21
6	4000 ²	90	74	30

¹Diet contained 45% cured meat as protein source.

²Only treatment in which NDMA was found.
Olsen and Meyer (1976).

TABLE 3.5
NITROSOPYRROLIDINE AND TUMOR FORMATION

Daily Dose mg/kg	No. Rats	With Tumors		Time of Death, Days
		Malignant	Bening	
0	61	6	5	587
0.3	60	12	8	664
1.0	62	20	9	685
3.0	38	32	3	533
10.0	24	11	6	444

Preussmann *et al.* (1976).

reported the presence of cancerous lymphomas in 12.6% of all animals receiving nitrite. Although 8.5% of the control animals also developed lymphomas, the difference between the treated and control groups was statistically significant. Newberne concluded that "... the data are suggestive and the biological significance of nitrite-associated lesions of the lymphoreticular system is unclear." There has been considerable scientific criticism of the procedures used in the experiment and the evaluation of the data. A committee has been appointed to review the histological sections; their work is not completed at this time. The Food and Drug Administration and the Department of Agriculture have requested a decision from the Justice Department on the legality of phasing out the use of nitrite under the Delaney Amendment, based on the validity of the Newberne Report.

EFFECT OF A BAN ON NITRITE

Nitrite is unique as a curing agent, affecting the flavor, color, texture, and rate of lipid oxidation of cured meat products. Its most important effect, however, is its inhibition of the outgrowth of and toxin formation by *Clostridium botulinum*, thus preventing the occurrence of botulism, a potentially lethal food poisoning condition. Prohibition of the use of nitrite would increase the risk of botulism from cured meat products. Besides the health hazard, banning nitrite would pose an economic problem. Economic impact analyses were published by the Council for Agricultural Science and Technology (CAST) (1978), Department of Agricultural Economics, Purdue University (Brandt *et al.* 1978), and the Economics, Statistics and Cooperative Service (ESCS), USDA. These are necessarily estimates based on multiple assumptions, so they only indicate the possibilities that might occur. CAST (1978) assumed that all bacon production would stop and there would be no utilization of the pork bellies; the industry would lose a \$1.5 billion market. The additional loss of medicinal byproducts and the effects on ancillary industries would raise the total loss to many billions of dollars. The Purdue analysis (Brandt *et al.* 1978) assumes that other meat products, principally sausage and luncheon meat, would absorb the pork bellies not used for bacon. In 1977 more than \$5 billion was spent on bacon, sausage, and luncheon meat, and if all of the bacon disappeared, there would be a direct loss of \$2.57 billion. Using all of the excess pork bellies to make more sausage and luncheon meat would result in a return of \$4.92 billion; the industry would lose \$110 million. However, if only 2/3 of the bellies were used for the substitute products, the report forecasts a sale of \$3.16 billion, or a loss of \$1.87 billion. These losses were assumed to be short-term only; adjustments would be made for the long-term, including fewer hogs produced by fewer producers, closing of some slaughter facilities, and a reduction in the number of employees in the chain. Effects would be felt in the distribution and retail sectors as well, and consumer prices would rise. ESCS (1978) has two scenarios in its analysis. If 50% of the bellies were used for nitrite-free bacon and the remainder processed into sausage, lard, and other products farm income would drop \$600 million and the Consumer Price Index (CPI) would go up 0.3%. If only 15% of the bellies were used for bacon substitutes, income would drop about \$1.3 billion and the CPI would increase 0.7%. Large quantities of available lard would depress soybean and other oilseed markets, and the amount of animal fat in the human diet would increase. Over a 5 year period, however, hog prices would stabilize at about 2% below baseline, fewer animals would be produced, and meat prices would be higher.

The effects of a partial ban of nitrite and nitrate in some meat products

can be seen in Norway. Instituted in 1973, the ban has not been complete, and some products are permitted with as much as 200 mg/kg nitrite. Hoyem (1976) reported that: sales were reduced about 5% initially but soon returned to normal; gray colored products, labeled with a statement that no nitrite was added, had to be withdrawn from the market; the consumer preferred the pink product when a choice was available. Producers developed some procedures to induce color formation, including: cooking and smoking products without nitrite in the same chamber as products legally containing nitrite, adding nitrite salts to sawdust prior to combustion, storing raw sausages for a few hours to get microbial reduction of natural nitrate, and increasing the nitric oxides in the air in the smokehouse by such means as burning propane gas. There is no serious concern about the shelf-life of the product, and no cases of botulism have been reported even from the consumption of products from which nitrite was excluded.

CURRENT STATUS

In 1973 the Secretary of Agriculture appointed an expert panel to review the risks and benefits of nitrite and nitrosamines in cured meat products. At the expiration of its charter in 1977, the panel presented a number of recommendations. Nitrate should be eliminated from all cured meat products except dry cured hams and Lebanon bologna, for which the concentrations permitted were to be reduced drastically. The concentrations of nitrite permitted were to vary with the class of product, but in all cases were to be lower than in use at the time. Since bacon is the major cured meat in which nitrosamines were found consistently, the Department acted on it first. Regulations issued set an ingoing target concentration of 120 mg/kg nitrite and a requirement for 550 mg/kg sodium ascorbate or erythorbate. A limit of 10 $\mu\text{g/kg}$ of NPYR in fried bacon was also established, with an action limit of 17 $\mu\text{g/kg}$. The Department recently began surveillance of commercially-produced bacon. Of the first 143 samples tested, 27 were out of compliance. However, modifications made in the processing procedures permitted a number of the processors to cure bacon that, after frying, had NPYR concentrations within the accepted limits.

Further reductions in nitrite concentration have been proposed by the Department. Unless otherwise directed, in May of 1979, bacon will be cured with 40 mg/kg nitrite, 0.26% potassium sorbate, and 550 mg/kg sodium ascorbate or erythorbate. This formulation is based on the claim by Ivey *et al.* (1979) that it will inhibit the outgrowth of *C. botulinum* to the same extent as the earlier-used 156 mg/kg nitrite alone. Considerable research is being conducted in essentially crash programs to evaluate the

effect of low levels of nitrite and sorbate on the various characteristics of cured meat products usually attributed to the higher concentration of nitrite.

Other substitutes for nitrite or inhibitors of nitrosamine formation have been suggested. α -Tocopherol was shown (Table 3.6) effectively to reduce nitrosamine formation in fried bacon, presumably because it is more fat soluble than ascorbate and erythorbate, and since NPYR has been shown to be formed principally in the adipose tissue. There is commercial interest in preparing bacon with this compound. A patent has been issued for the use of *tert.* butylhydroquinone and parabens (Sweet 1975) as nitrosamine inhibitors, and Rubin has discussed the effect of ascorbyl acetals for this purpose (1977). Federal meat inspection regulations have been amended recently to permit the use of lactic acid starter cultures, or cultures of *Pediococcus cerevisiae*, in bacon on the premise that the increased acid would reduce the concentration of residual nitrite and, thus, the concentration of nitrosamine formed (Anon 1979). Considerable research has been carried out in our laboratory on the inhibition of *C. botulinum*, and a number of compounds were found to be considerably more effective than nitrite in test tube studies but they were inactivated when used in bacon.

TABLE 3.6
 α -TOCOPHEROL AND NITROSOPYRROLIDINE FORMATION IN
BACON

Company	NPYR by GC-TEA (ppb)			
	A	B	C	D
1	8.9	15.8	1.4	0.5
2	19.5	13.2	10.1	8.2
3	6.5	3.9	3.9	0.5
4	1.8	0.5	0.5	0.5
5	<0.5	0.5	0.5	0.5
6	7.3	4.2	0.6	0.5
7	1.5	0.5	0.5	0.5
8	15.2	8.9	2.9	1.3
Mean \pm SD	7.58 \pm 6.88	4.5 \pm 4.75	2.36 \pm 3.45	1.25 \pm 2.87
NO ₂ (ppm)	200	120	200	120
ASC (ppm)	550	550	550	550
α -TOC (ppm)	0	0	500	500

The formation of nitrosamines in bacon could be avoided by the elimination of nitrite from the cure, but the risk of growth of

C. botulinum, which is an unknown factor, requires some action to reduce the possibility of botulism. It has been suggested that nitrite-free products can be maintained in a frozen condition to prevent the growth of the microorganism.

CONCLUSION

Recent demonstrations of the ubiquity of nitrite and nitrosamines, both in the environment and within the human body, suggest that now, more than ever, further information is needed about the role of these compounds in the concentrations in which they are being found in nature and their relationship to the carcinogenic process in humans.

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